VOLUME L NUMBER 5

BOTANICAL GAZETTE

NOVEMBER 1910

THE PEG OF THE CUCURBITACEAE

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 140

WILLIAM CROCKER, LEE I. KNIGHT, AND EDITH ROBERTS

(WITH SIX FIGURES)

Historical

The peg of seedling Cucurbitaceae has frequently been used as a marked case of adaptation to a peculiar function. It holds the seed coat while the elongating arms of the arch of the hypocotyl withdraw the cotyledons from the coat. The method by which the peg functions is well shown in fig. 1.

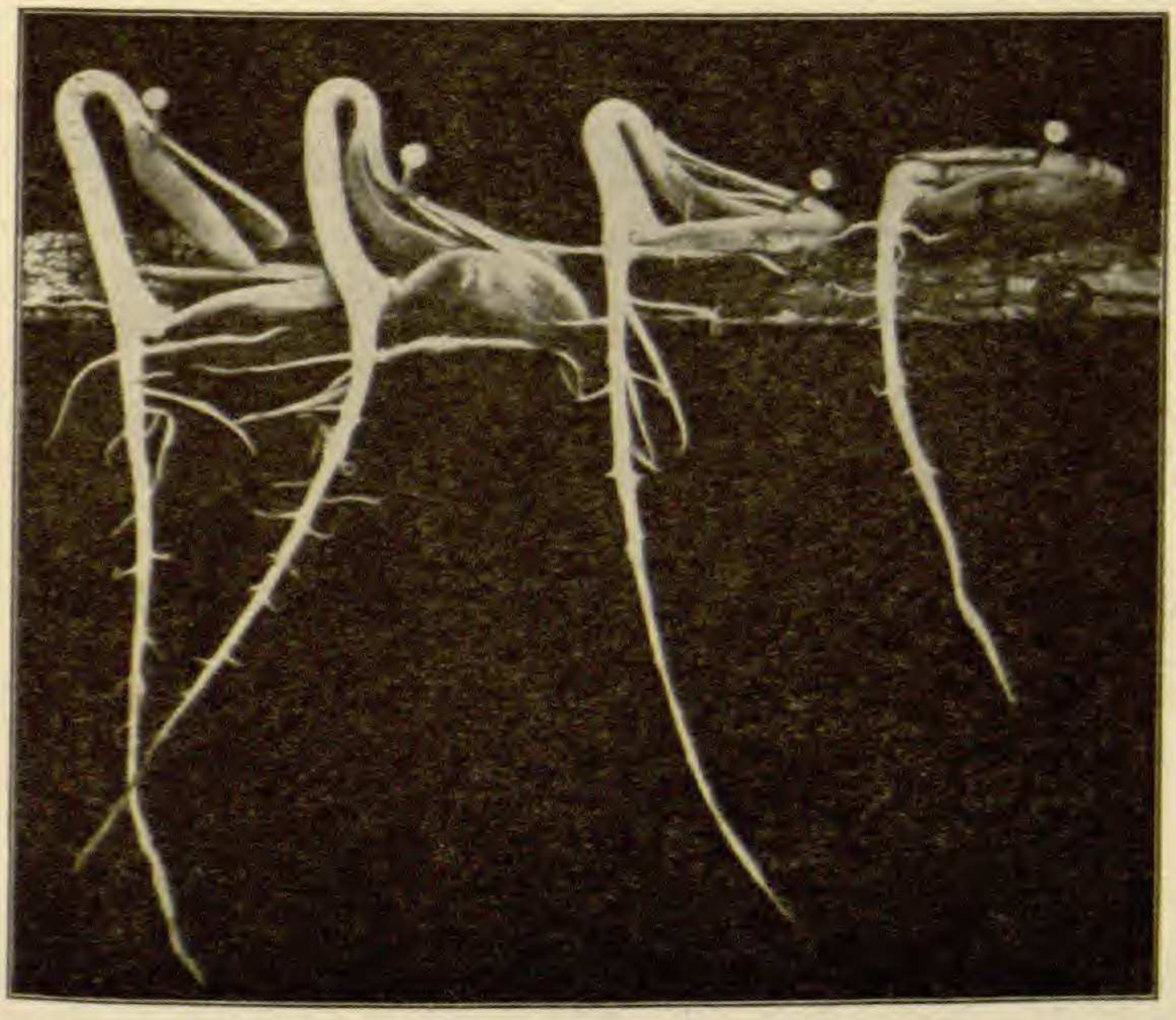

FIG. 1.—Big Tom, showing the pegs functioning in the removal of the coats.

MIRBEL[1] and TITTMAN[2] very early showed the presence of this organ throughout the Cucurbitaceae. While it is well developed in the epigean forms, it is very rudimentary in the hypogean forms, such as *Megarhiza californica* and *Sicyosperma gracilis*. The organ is by no means limited to the Cucurbitaceae, but appears in various genera of a number of families: *Mirabilis, Oxybaphus*, and *Abronia* (Nyctaginaceae), *Martynia* (Martyniaceae), *Lindheimera* (Compositae), *Mimosa* (Leguminosae), *Tribulus* (Zygophyllaceae), *Eucalyptus* (Myrtaceae), *Cuphea* (Lythraceae), etc. While in Cucurbitaceae the peg appears only after germination has progressed considerably, in other forms, as *Eucalyptus* and *Cuphea*, it is already laid down as a complete ring in the mature seed, and with germination completes its development.

TITTMAN recognized the biological significance of this organ in the Cucurbitaceae, and showed that it appears only on the lower side of the developing hypocotyl.

TSCHERNING[3] gives a rather full description of its histology and physiology. He states that it is a parenchymatous outgrowth. While the greater diameter of the parenchymatous cells in other parts of the hypocotyl is longitudinal, in the peg zone it is radial. The number of layers of cortical cells is also somewhat greater at the peg zone. TSCHERNING describes the effect of the position of the seed during germination upon the development and functioning of the peg. He says that when the radicle points vertically downward the swelling does not occur, and the cotyledons push above the ground still bearing the coat. With radicle pointing vertically upward, the peg develops on the concave side of the arch, but does not remove the coat. When the seeds are planted on edge, the peg develops on the concave side of the arch and wedges between the two valves of the coat, thus succeeding in removing a considerable percentage of them. When the seeds are planted on a flat face, the peg develops on the concave side of the arch and attaches itself securely to the lower valve of the coat, thereby insuring its removal. TSCHERNING emphasizes the development of the peg on the concave side of the arch, and speaks of it as a lateral pushing out of the cells due to the inhibition in the

[1], [2], [3] See under NOLL, footnote 8.

elongation on the concave side of the hypocotyl. While his relating its place of development entirely to the arch is probably correct, as we shall see later, his view as to how the arch brings about such a placement is crude. We shall see also that his claim that the peg does not develop when the seed grows with the radicle pointing vertically downward is incorrect.

FLAHAULT[4] claimed that seedlings that developed when the removal of the coats occurred in the normal way were far superior to those in which it was prevented by breaking away a portion of the lower valve of the coat so that the foot could not get a hold. This advantage he attributes of course to the hindering of assimilation by the retained coats. He claimed that the peg develops at any point on the hypocotyl necessary to enable it to hold the coat.

CHARLES DARWIN,[5] contrary to FLAHAULT, found that the peg develops only at the zone between root and stem. The lower face is root, as shown by the presence of root hairs and reaction to potassium permanganate, while the upper face is stem. DARWIN described a number of experiments, similar to those of TSCHERNING, testing the effect of the position of the seed upon the development and functioning of the peg. While DARWIN clearly showed that the peg is located at the border between root and stem, he did not show the stimuli involved in its lateral placement.

FRANCIS DARWIN[6] later showed that seedlings of *Cucurbita ovifera*, allowed to germinate on a slowly rotating clinostat with a horizontal axis, gave pegs completely surrounding the hypocotyl and approximately equal on all faces. He concluded, from the inadequate experiments of his father and himself, that gravity is a direct stimulus determining the lateral distribution of the peg, and that therefore this experiment shows that gravity is continually effective on the clinostat and only equalized in its action on the several flanks of the exposed object. The conclusion concerning

[4] FLAHAULT, C., Sur le talon de la tigelle de quelques Dicotylédones. Bull. Soc. France **24**: 200. 1877.

[5] DARWIN, CHARLES and FRANCIS, The power of movement in plants. pp. 102–107. New York. 1892.

[6] DARWIN, F., and ACTON, E. H., Practical physiology of plants. pp. 192 ff. Cambridge. 1895.

the nature of the effect of the clinostat is correct, as shown by FITTING[7] and others, but the assumption that this experiment proves it is quite another question, and, as we shall show later, quite out of accord with a number of other facts.

NOLL[8] mentioned the fact that FRANCIS DARWIN'S conclusion that gravity is a direct stimulus in determining the lateral distribution of the peg is not proved by his meager experiments. He also claimed that only 50 per cent or less of the seedlings grown on a clinostat produce ringlike pegs, while the remainder show sharp arching, with a one-sided peg. NOLL stated that all flanks of the hypocotyl are qualitatively equally capable of peg-development, but that quantitatively the broader flanks produce the larger peg. He concluded that contact of the seed coats is not a necessary stimulus to peg-development, for the peg still appears when the coats are removed. The contact of the coat, however, increases the size of the peg. He performed a number of experiments in which seeds were germinated with the two opposite broad faces exposed to gravity alternately and for equal periods. In all these cases the two faces developed essentially equal pegs. He especially emphasized these as showing that the peg stands in strong relation to gravity and owes its stimulus for formation to gravity. Many experiments were also performed in which seedlings were grown with the radicle directed perpendicularly downward or at a slight deviation from this position. From these he concluded that the gravity stimulus reaches beyond the lower pole of the seedling axis. At a deviation of 5° from the vertical he found a weak peg on the upper side, while at 7°5 the peg was almost exclusively on the lower side. He fixed the limit of the field of stimulation at 6°–8° deviation from the vertical. He specified that these are to be taken only as approximate figures and as applying to *Cucurbita Pepo*. In two seedlings slight pegs were found on the upper side at a deviation of 8°. Seedlings were also found that showed no peg, but he did not know how to account for this. On account of great variation in response, NOLL empha-

[7] FITTING, HANS, Jahrb. Wiss. Bot. **45**:575–600. 1909.

[8] NOLL, F., Zur Keimungs-Physiologie der Cucurbitaceen. Landwirt. Jahrb. **30**:145–165. 1901.

sized that reliable conclusions can be drawn only by the employment of a large number of seedlings in each culture.

It is evident that both FRANCIS DARWIN and NOLL considered gravity as well as the arching of the hypocotyl as direct stimuli in determining the lateral distribution of the peg. NOLL says (p. 164): "Die meiste inseitige Ausbildung des Wulstes tritt als Ergebnis zweier heterogener Reize ein. Die localisierte Entstehung des Wulstes ist einerseits abhängig vom Gravitationsreiz. Der Wulst bildet sich auf der jeweiligen Unterseite. . . . . Die einseitige Wulstbildung wird andererseits auch bedingt durch die Krummung des Mutterorgans, derart, dass auf der Konkavseite die Bildung des Stemmorgans ausgelöst wird."

FRANCIS DARWIN[9] has lately used the peg of the cucurbits as a mainstay for the memory theory of plant response. In this, of course, he assumed that its development and position are directly determined by gravity. Whether gravity acts as a direct stimulus to its production and placement is the principal question to be tested by the following experiments.

Methods and materials

NOLL failed to control the factors involved in this problem in a way that enabled him to determine the part played by each in the lateral placement of the peg. He apparently failed to notice the potency of contact of the coats on the one hand, and of the media on the other, in arch-production. In the experiments here given, to avoid this important influence, except when its effect was to be studied, the seeds, whether on the clinostat, centrifuge, or in rest positions of different inclinations, were freed from the coats at the tips and each held by two pieces of cork between which the cotyledons were clasped at the central region. The whole apparatus was kept in the dark and watered by a very fine spray heated to 23° C. The spray was formed by forcing the water by means of tap pressure through a tank of considerable size kept in a water bath at constant temperature, and allowing it to break against a plate of glass. In this way contact of both the coats and the soil media is entirely eliminated. Contact, as data later given will

[9] DARWIN, FRANCIS, New Phytologist 5:199–207. 1906.

show, is a very important influence. We are unable to see how NOLL could get results at all dependable without taking this precaution. He was always studying the effect of two stimuli when his results purport to be considering one.

The seeds used in this work were obtained from VAUGHAN and will be mentioned by the trade name used by that dealer.

Experiments and discussion

EFFECT OF CONTACT OF COATS

As has been stated, the contact of coats plays a very important part in arch-production, and therefore indirectly upon the lateral

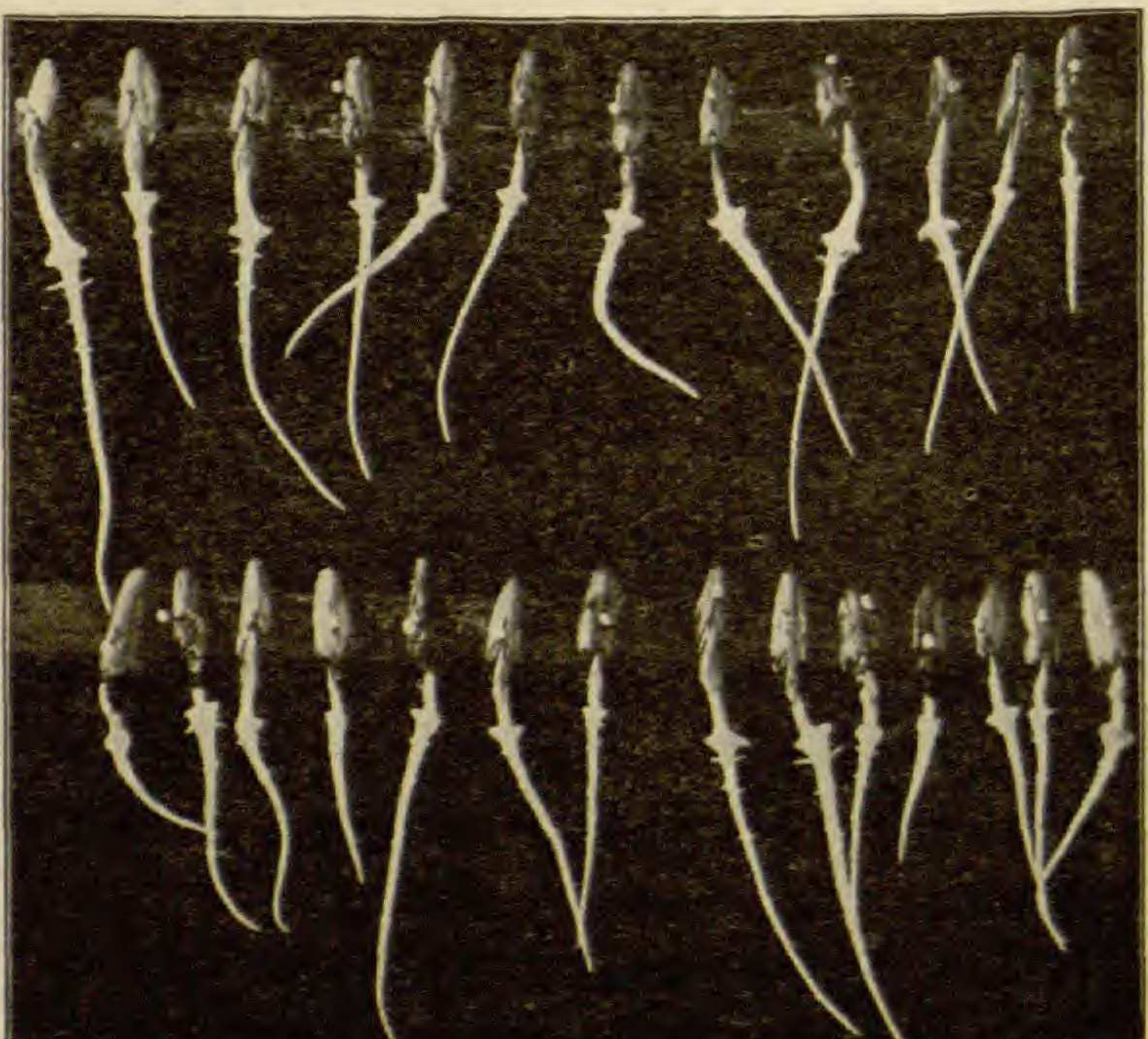

FIG. 2.—Crop of Hubbard squash grown in spray at 23° C.; coats removed at the tip and radicles pointing downward.

placement of the peg. This effect was tested in two ways: (1) by growing seeds upon a clinostat with coats intact and with coats removed at tips, and (2) by a similar growth of seeds held between cork strips with radicles pointing downward or approximately so. These were kept in a dark chamber and watered with a spray at 23° C., as described above. There is some variation in different varieties in the response to contact. For example, the pumpkin called Big Tom, especially when grown on the clinostat, gives a somewhat higher percentage of strong arching in response to coat contact than does the Hubbard squash. Fig. 2 shows all the individuals of a culture of Hubbard squash grown as described above, coats removed at tip, and radicles pointing downward. Of the 26 seedlings, none are sharply arched, and all have pegs either two-sided or ringlike. Three show slightly more prominent pegs on one side than on the other. It is evident then that in

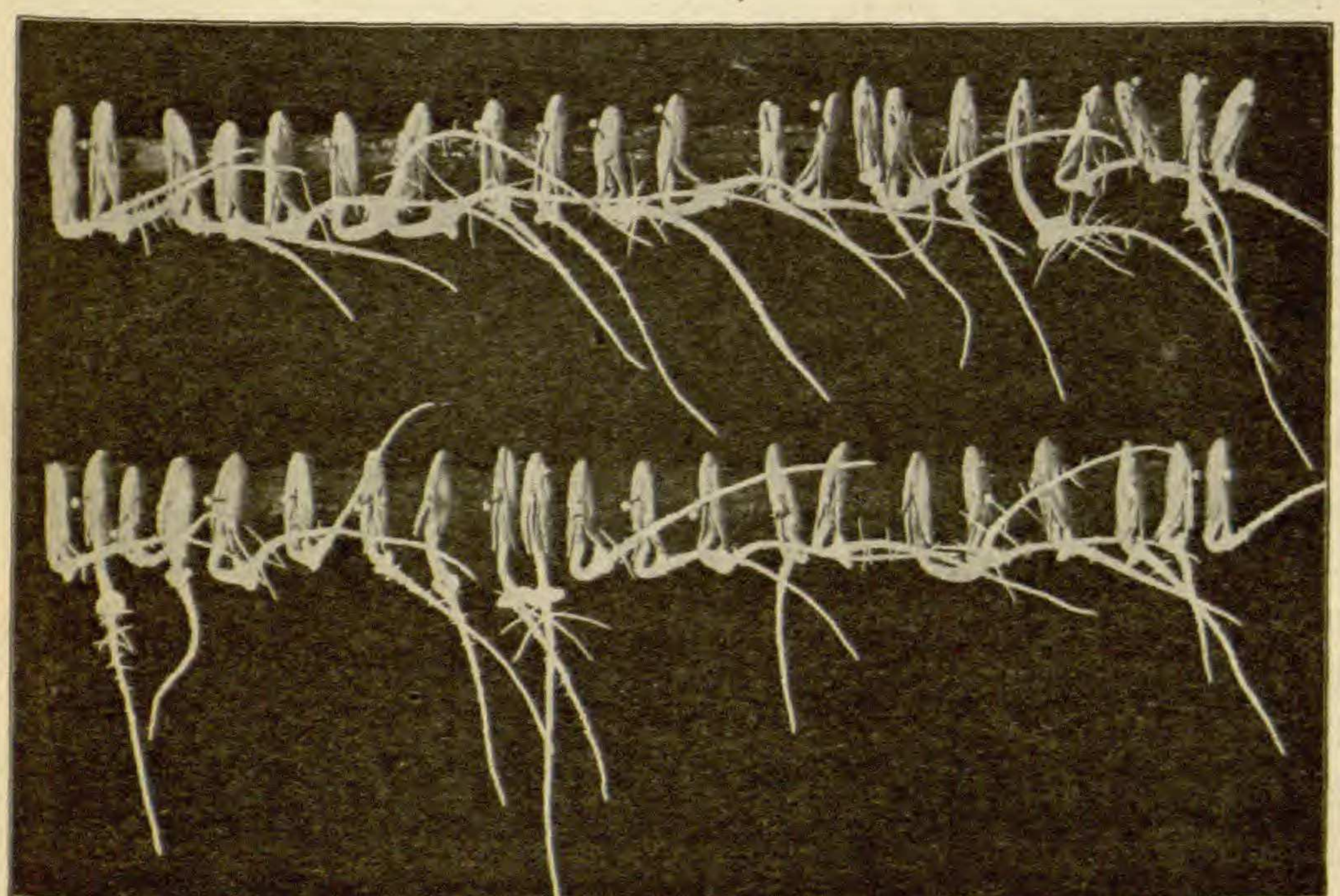

FIG. 3.—Crop of Hubbard squash as in fig. 2, but coats not removed at tip; shows sharp arching induced by contact of coats.

the Hubbard squash little arching and no one-sided pegs are produced if the seedlings grow with the radicles pointing vertically downward, and with no contact of either coat or media. Fig. 3 shows a similar culture except that the coats were intact during the growth. Of the 42 seedlings, 29 are sharply arched and 13 very slightly or not at all. This shows contact of coats to be very potent in arch-production. Of those that are sharply arched, 8 have pegs entirely on the concave side of the arch, while several others show the greater peg-development on that side. In this case the arching is against gravity, which emphasizes the potency of contact of coats as a stimulus to arching. By a comparison of figs. 2 and 3, one can see clearly the effect of contact upon the size of the peg.

A culture of pumpkin (Big Tom), with coats removed, radicles pointing vertically downward, and grown in a spray as described above, produced 39 seedlings. Of these none were sharply arched; 19 showed very small two-sided pegs; 19 were essentially pegless; and 1 showed a small one-sided peg on the concave side of the slight arch. A similar culture of the pumpkin (Big Tom) with coats intact gave 31 seedlings; of these, 25 were sharply arched, with pegs entirely on the concave side of the arch; 1 was little arched, with peg on concave side of arch; 3 were pegless; and 2 had equal, small, two-sided pegs. From these data it is evident that in the pumpkin (Big Tom) contact of coats is even more effective in producing arching than in the squash (Hubbard), and unlike the case of the squash, the arching shifts the peg entirely to the concave side of the arch. From the large number of pegless seedlings appearing in the culture with the coats removed, it is also evident that the arch not only determines the lateral placement of the peg, but in some cases even its appearance. One may be inclined to think the existence of the peg is perhaps determined by the contact of the coat, rather than by the arching produced by the coat; but it is not probable, for of the 6 not arched in the culture with coats on, 3 are pegless. It is apparent, therefore, that a large number of the pumpkins (Big Tom) are pegless if developed without considerable arching, and that arching determines in about half the cases whether or not there will be a peg, as well as the placement of the peg on the concave side of the arch.

Fig. 4 shows a culture of pumpkin (Big Tom) with coats removed at the tips, and grown on the horizontal clinostat in a spray. Of the 30 seedlings in the culture, none are sharply arched; 14 have small two-sided pegs; 8 show slight pegs on the concave side of the slight arch; and 8 are essentially pegless. Fig. 5 shows a similar culture except the coats are intact. Of the 24 seedlings, 20 are sharply arched, with pegs entirely on the concave side of the arch; 2 are slightly arched, one of which has an equal two-sided peg and the other a slight one on the concave side of the arch;

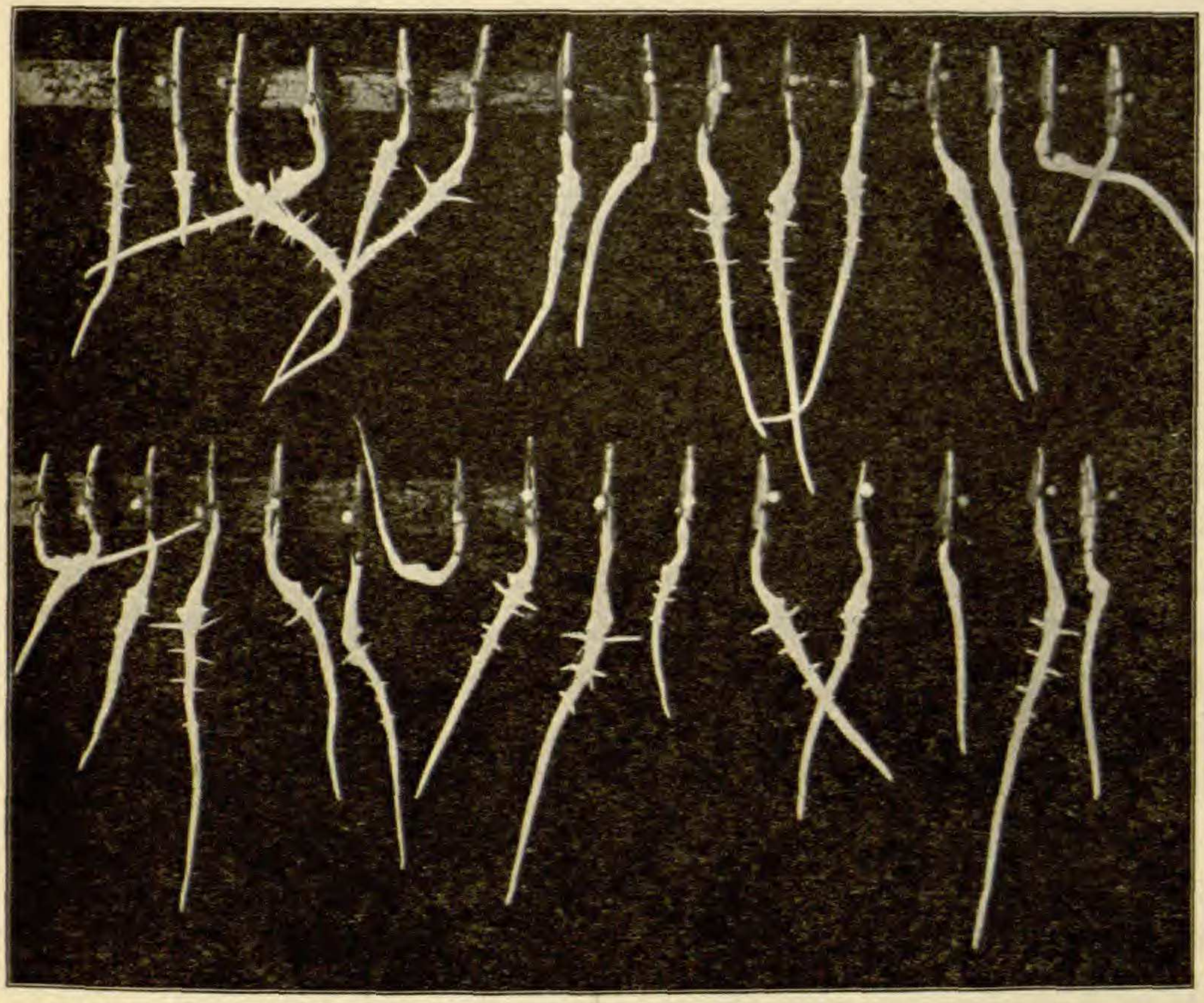

FIG. 4.—Crop of Big Tom grown on a clinostat in a spray; temp. 23°; coats removed at the tip.

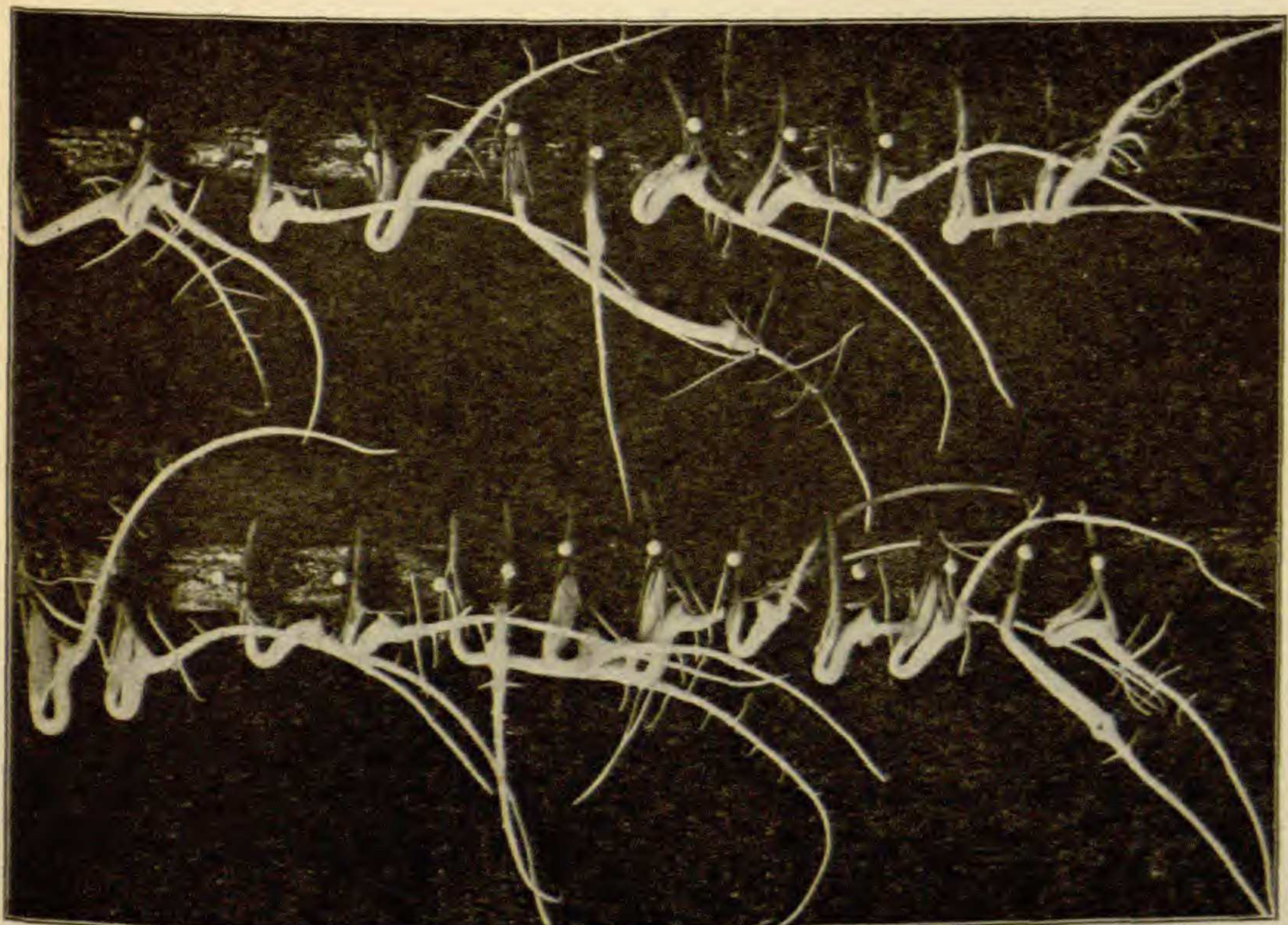

FIG. 5.—Crop of Big Tom grown as fig. 4, except coats were not removed at the tip.

2 show no arch and are essentially pegless. One is struck here by the small percentage of pegless seedlings in the culture in which the coats are removed, as compared with corresponding seedlings grown with the radicle pointing vertically downward. This is easily explained, however; the arching does not have to take place against gravity, due to the equalization of this stimulus by the clinostat. In short, the autonomic disposition to arch is unrestrained. This fact, with the fact that slight arching produces a one-sided peg in this form, makes the data exactly what must be expected. Again, one sees that the contact of the coats is much more effective in producing arching and one-sided peg-development than in the culture in which the seedlings developed with the radicle pointing vertically downward. This again must be expected, for the arching does not have to occur in opposition to gravity, since this stimulus is equalized by the clinostat.

NOLL states that more than 50 per cent of pumpkins grown on a horizontal clinostat show sharp arching, with the peg entirely on the concave side of the arch. This is the case if one allows two factors to act at once: contact of coats and the autonomic tendency to arch. But NOLL's conclusion that such results will follow from the autonomic tendency to arch alone is erroneous, and was possible only because he overlooked the very important influence of contact of coats in the arching. DARWIN's statement that in seedlings of *Cucurbita ovifera*, grown on the clinostat, the peg is almost equal on all sides, certainly holds for the squash (Hubbard), providing both contact of coats and media are avoided, as they were in our clinostat cultures.

From the experiments stated in this section several things are evident. Contact of coats is an extremely important stimulus for arch-production, both in the pumpkin and the squash. It produces strong arching even against gravity. In the pumpkin (Big Tom) any considerable arching increases greatly the percentage of seedlings that show pegs; and it causes the development of the peg on the concave side of the arch. In the squash (Hubbard) arching is far less effective in determining the lateral placement of the peg. The autonomic tendency to arch is very slight, though it tends to a lateral placement of the peg in the pumpkin (Big

Tom), as shown by the clinostat experiments where the coats are removed. It is also evident that results obtained without reference to the effect of contact of the coats cannot lead to a knowledge of the part played by the several factors in determining the lateral placement of the peg.

EFFECT OF DEVIATION FROM THE VERTICAL POSITION

As has been stated, NOLL claimed that seedlings grown in a vertical position, with the radicle pointing downward, gave two-sided or ringlike pegs. He found, however, that a slight deviation from this position (8°5) produced only one-sided pegs. We ran numerous experiments on various varieties of cucurbits for the purpose of testing this statement. The results are tabulated in the accompanying table. "Vertical" in this table means that the seed was held so that the plane passing through the largest dimension of the seed was in a vertical position and the radicle was pointing downward. Deviations from this plane were made by tilting the plane of the seed the number of degrees from this position that is recorded in the table. The tilting always exposes one face rather than the edge of the seed to gravity. The seeds were held in position by two pieces of cork lightly clasping them at the central region. The whole culture was grown in darkness in a spray, so that no contact of media was involved and no contact of coats except when specified.

A glance at this table makes it very evident that it furnishes no ground for NOLL's conclusion that a deviation of over 8°5 gives only one-sided pegs. Our extensive experiments make us unable to understand how he obtained such results.

It is evident from the table that in every variety used the deviation from the vertical is far more effective in the lateral placement of the peg when the coats are intact than when they are removed at the tip. This is due to the fact that the arching is made much sharper by the coat contact, and the lateral placement is in turn furthered by the sharper arching.

It is also noticeable that there is a great variation in the several varieties in the effectiveness of the deviation for the lateral placement of the peg. In Big Tom it is most effective, while in Boston

Deviation from vertical position		Vertical			10°			20°			30°			45°			65°			90°			135°			150°			165°			180°		
Nature of peg																																		
Variety	Condition of coat at tip	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless	1-sided	2-sided	pegless
Big Tom (pumpkin)	removed	6	27	37	0	4	5	1	6	6	0	11	4	16	27	5	18	6	0	47	19	0	59	0	0	60	0	..	57	0	..	42	0	..
	intact	30	7	1	15	1	0	16	4	1	3	3	..	35	4	0	17	0	0	43	1	0	101	0	1	30	0	..	46	0	..	50	0	..
Hubbard (squash)	removed	5	44	1	0	8	0	3	10	..	..	..	..	22	41	0	..	..	..	38	3	0	38	3	0	..	..	..	..	..	..	..	..	..
	intact	3	23	1	1	10	0	1	12	..	..	..	..	27	17	0	..	..	..	46	2	0	46	2	0	..	..	..	..	..	..	..	..	..
Boston marrow (squash)	removed	..	..	..	..	..	..	22	22	..	1	27	..	0	6	0	5	15	..	..	..	..	19	9	0	24	8	0	54	3	0	58	4	..
	intact	..	..	..	6	29	..	6	42	..	13	11	..	..	..	..	8	4	..	..	..	..	35	2	0	38	5	0	47	4	0	45	1	..
Calhoun (squash)	removed	..	..	..	..	..	..	..	..	..	0	23	..	2	13	0	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..
	intact	..	..	..	..	..	..	..	..	..	2	13	..	4	5	0	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..
Sugar pie (pumpkin)	removed	..	..	..	..	..	..	..	..	..	8	2	7	27	31	4	..	..	..	103	29	12	..	..	..	..	..	..	..	..	..	..	..	..
	intact	..	..	..	..	..	..	..	..	..	9	3	9	23	4	3	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..	..

marrow it is least so. In the former no two-sided pegs appear at 135° deviation or above, while the latter at 180° deviation still shows about 8 per cent with two-sided pegs.

In the exposures with the coats removed, one can see the effectiveness of gravity alone in the one-sided peg-production. It must not be forgotten that the effect of gravity in lateral placement of the peg is probably entirely indirect and acts through the production of an arch.

As has been mentioned, NOLL thought that not only the development of the peg but its lateral placement is called forth by the joint action of two heterogeneous stimuli: gravity, which causes the development on the lower side of the hypocotyl; and "organform" stimulus, which causes the development on the concave side of the arch. NOLL compared the second of these to the appearance of lateral roots on the convex side of a curved main root. His main evidence for the conclusion concerning two heterogeneous stimuli is based on his experiments in which he grew seedlings at various deviations from the vertical (radicles pointing downward). If his statement that any deviation beyond 8°5 always produces a one-sided peg were true, his conclusion would be entirely justified. Our results, derived by the use of five different varieties of cucurbits, of very different characters so far as peg-development is concerned, and obtained with greatest care in eliminating all factors, except gravity, show that it takes a deviation of not merely 8°5, but in every case of more than 90°, and in forms like the Boston marrow of more than 135°, to insure a one-sided peg. When a seedling grows in the horizontal position, gravity and the "organform" stimulus certainly are both acting to produce the peg on the lower side only, and yet our results show that many seedlings thus grown produce two-sided pegs. NOLL's two heterogeneous stimuli are not adequate to explain these results. This view becomes yet less in accord with fact when it is remembered that NOLL assumed that gravity could not reach more than 8°5 over the lower pole of the hypocotyl, that is, that gravity is not effective in inducing peg-development on the upper side of the hypocotyl if the deviation from the vertical is over 8°5.

It has already been pointed out that in *Eucalyptus* and *Cuphea* a ringlike peg is laid down in the formation of the seed, and with germination this enlarges somewhat. In these forms, therefore, the peg is a natural outgrowth approximately equal on all flanks of the seeds; its appearance and development are related, undoubtedly, to the plant form and not called forth alone by external stimuli. We find that such a ringlike peg, rather small but approximately equal on all flanks, develops on the seedling of palo verde (green-stemmed *Parkinsonia*). In this form the arching of the hypocotyl is not marked and the peg does not function in the removal of the coat; likewise, external conditions do not seem to lead to a lateral placement. Results already stated, as well as data to be stated later, lead us to believe that the peg of the cucurbits is, to a considerable degree, a natural outgrowth of the seedling, and that it is approximately equal on all flanks if arching is avoided. We must of course accept the quantitative greater development on the two broader faces of the hypocotyl over the two narrower. It is also evident that in different species the size of the outgrowth varies greatly; it is relatively large in squashes (Hubbard, Boston marrow, and Calhoun), and relatively small or even absent in pumpkins (Big Tom and Sugar pie). We have also found the latter to be the case in various watermelons and cucumbers. In fact, if arching is avoided, different species will show all the intermediate forms from the so-called pegless (fig. 6) to those that have very prominent ringlike pegs. We must also recognize that the contact of coats, as figs. 2 and 3 show, greatly increases the size of this outgrowth. The size is also greatly increased, at least on one flank, by arching.

FIG. 6.—Pegless seedlings of Big Tom.

The question, therefore, does not seem to be so much what stimuli cause the development of this organ, as DARWIN and NOLL assumed, but rather what stimuli lead to its lateral placement and tend to increase its size.

It is certainly evident, as all who have worked on this subject agree, that arching or "organ-form" stimulus, as NOLL termed it, leads to the shifting of the peg to the concave side of the hypocotyl, or, as it would be better termed, to the development of the peg on the concave flank of the hypocotyl. Both gravity and contact of coats aid in arch-production. Of these, as our results show, the latter is much more important. Contact of coats also greatly increases the size of the peg. Other questions that should be answered in this connection are whether or not gravity acts as a direct stimulus in the lateral placement of the peg, and whether or not it tends to increase the size of the peg on the flank exposed to it.

As has been repeatedly said, both DARWIN and NOLL assumed that gravity is a direct stimulus, not only in determining the existence of a peg but also its lateral placement. DARWIN'S evidence for this was that a ringlike peg is found when the seedlings of *Cucurbita ovifera* were grown in a horizontal position on a slowly rotating clinostat. NOLL'S evidence for this conclusion included two other facts: (1) seedlings with the radicle pointing downward but deviated more than 8°5 from the vertical gave pegs only on the lower side, and (2) seedlings turned over every few hours during development gave pegs ringlike or two-sided. We have shown that NOLL'S deviation experiments do not at all accord with results obtained with all care to avoid the variation of more than one factor at a time. As for the clinostat and turning evidence, it is just what would be expected if the peg is an outgrowth approximately equal on all sides, if sharp arching is avoided. Of course the clinostat and the turning from time to time prevent sharp arching, and thereby give the natural ringlike or two-sided development.

Further evidence as to whether gravity is a direct stimulus in determining the existence of a peg, its lateral placement, or its size, is found in the sections that follow.

CENTRIFUGE CULTURES

A large number of cultures of the several varieties of cucurbits worked with were made on a centrifuge with a vertical axis. The acceleration varied from two gravities to eighteen gravities. The cultures were grown in a spray at 23° C.; the coats were removed to avoid all contact; and the long axis of each seed was arranged parallel with a radius of the centrifuge, the radicle directed away from the axis.

Three facts were noticeable in such seedlings, especially in those grown with the greater centrifugal acceleration: (1) the hypocotyls were very straight, (2) the pegs were smaller than in similar cultures on clinostat or with radicles pointing vertically downward, and (3) the pegs though small were approximately equal on all flanks.

If, as NOLL assumes, gravity calls forth the peg in seedlings with the radicles pointing downward, one would expect larger pegs with increased gravities, unless the rather remarkable situation exists that one gravity is the optimum or is greater than the optimum for peg-development. But how can one account for the more meager and very regular peg? This is undoubtedly due to all arch strains being overcome by the centrifuge, which removed this stimulus to the enlargement and unequal distribution of the peg.

It must not be forgotten that a centrifuge thus operated gives two mass accelerations at right angles to each other: one due to the centrifugal acceleration produced by the machine and acting in the direction of the pointing root, and one gravity downward. If a flat side of the seedling faces downward, and the machine gives a centrifugal acceleration of three gravities, three gravities act in the direction of the pointing root and one gravity downward. Such cultures showed very slight tendency to arch, and no greater peg-development on the lower than on the upper flank. If this organ were sensitive to gravity, one would expect it to show a greater response on the side where one extra gravity was acting, especially when the centrifugal acceleration was only three gravities. Of course it is entirely possible that WEBER'S law applies in such proportions that the application of one gravity as a stimulus cannot be perceived when three gravities are acting at right angles to it. In geotropism of *Vicia Faba* epicotyls, however, WEBER'S law applies in the proportion of 24–25, that is 4 per cent excess exposure to gravity in one direction is sufficient to give a response.

It must be remembered that while this experiment does not prove that gravity is not a direct stimulus in determining the size and lateral placement of the peg, it certainly furnishes no evidence that it is.

A detailed statement of the results of one experiment with Big Tom on a centrifuge with a horizontal axis will suffice to indicate the nature of the results obtained with that apparatus. Here, also, the coats were removed at the tip and the temperature kept at 23° C. by means of a spray. The revolutions were 440 per minute and the circles 3 and 6 inches in diameter. The 3-inch circle gives 8.25 gravities, while the 6-inch gives 16.5. In the 3-inch circle 15 were essentially pegless and 2 with slight two-sided pegs. In the 6-inch circle 25 were essentially pegless, 4 with slight two-sided pegs, and one with a peg toward one edge of the seed. The marked tendency to show pegless forms is manifested here, as in all experiments on the centrifuge, with three or more gravities. It seems that these accelerations prevent all disposition to arching and to arching nutations; hence they give many pegless forms.

OBLIQUE CLINOSTAT CULTURES

A number of cultures were made on the oblique clinostat as devised by FITTING.[10] The cultures were watered by a spray at 23° C. and grown with coats removed at the tip to avoid contact. The end of the clinostat axis to which the frame for bearing the seeds was attached was pointed upward from the horizontal, while the radicles of the seeds pointed toward the axis. This insures, as an understanding of the oblique clinostat will show, that extreme arching is avoided. The axis was placed at various angles with the horizontal, and the large plane of the seeds at corresponding angles with the axis. When the axis varied 5° from the horizontal, and the large plane of the seed was broken 5° from the axis, at one point in each revolution one flat face of the seed was exposed to gravity at an angle of 80° from the vertical (radicle pointing downward) and the other face at 90°. As soon as considerable growth occurs, of course geotropic response occurs in favor of the 90° exposure, and this throws the flank of 80° exposure more nearly

[10] See footnote 7.

90°, thereby avoiding sharp arching. The difference of exposure is marked then only in the early stages, and other exposures with 90° against 76° and 90° against 85° gave no peg-development in favor of the 90° either in the pumpkin or squash. It has been shown that the effectiveness of geotropic stimulus in orthotropic organs is approximately proportional to the sine of the angle, and therefore the 90° exposure is stronger than any of the others. If gravity is effective as a form stimulus in causing the lateral placement of the peg, we might expect it to be manifested by a larger peg-development on the flank exposed at 90°. These experiments give no indication of it, and yet it would hardly be expected that such differences in exposure would show an effect, since much greater differences failed to do so in the centrifuge experiments described above. It must also be pointed out that, unlike the centrifuge experiments, these experiments are rather unsatisfactory because the arching occurs in favor of the 90°, and that factor is sufficient, if great, to produce lateral placement of the peg. On this account, only angles differing rather slightly can be compared. It is evident, however, that these experiments, as do the centrifuge experiments, furnish no evidence that gravity is a direct stimulus to the lateral placement of the peg.

General and summary

As one sees from the experiments given above, there is *no evidence that gravity acts as a direct stimulus to the lateral placement of the peg.* Certainly, then, DARWIN is not justified in using this assumption as a main prop to a theory (mnemic theory) which itself looks away from rather than toward progress in the knowledge of plant response.

Assuming that all arching is avoided, the following facts seem to hold: the peg is to a considerable degree a natural integral part of the plant; it develops on all flanks of the hypocotyl approximately equal (granting perhaps that it is somewhat larger on the broad flanks in many of the cucurbits); it varies in size from the very slightest outgrowth appearing in a small percentage of Big Tom to the large pegs of the Hubbard squash.

It may be laid down with the formation of the seed, as in

Eucalyptus and *Cuphea,* where it is an equal ring all around the organ; or its formation may begin after germination, when its position and size are determined by the factors we have shown to be effective.

The lateral placement is apparently brought about by the arching of the hypocotyl. Two stimuli aid in the formation of the arch: contact of the coats and gravity. Contact of the coats is by far the more effective, for it will induce very sharp arching even against gravity.

In forms like the Boston marrow, gravity independent of contact with a deviation of 170° from the vertical gives strong enough arching to produce only 90 per cent with one-sided pegs. In Big Tom and various other forms it is somewhat more effective.

Arching leads to an increased development of the peg, as well as to its lateral placement, and in many cases it produces a peg where it would not otherwise appear, as in Big Tom. Contact likewise increases the size of the peg independent of its effect through arch-production.

No evidence obtained from this detailed study indicates that gravity, as a direct stimulus, in the least increases the peg-development.

THE UNIVERSITY OF CHICAGO